Chapter 8

Ultrasound Application to Improve Meat Quality

Alma D. Alarcon-Rojo, Esmeralda Peña-González, Iván García-Galicia, Luis Carrillo-López, Mariana Huerta-Jiménez, Raúl Reyes-Villagrana and Hector Janacua-Vidales

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http://dx.doi.org/10.5772/intechopen.77973

Abstract

High-intensity ultrasound offers an alternative to traditional methods of food preservation and is regarded as a green, versatile, popular, and promising emerging technology. Ultrasound generates acoustic cavitation in a liquid medium, developing physical forces that are considered the main mechanism responsible for the observed changes in exposed materials. In meat, ultrasound has been successfully used to improve processes such as mass transfer and marination, tenderization of meat ,and inactivation of microorganisms. It is also an alternative to traditional meat aging methods for improving the quality properties of meat. Moreover, the combination of ultrasonic energy with a sanitizing agent can improve the effect of microbial reduction in foods. This review describes recent potential applications of ultrasound in meat systems, as well as physical and chemical effects of ultrasound treatments on the conservation and modification of processed meat foods.

Keywords: ultrasound, cavitation, emerging technology, minimal processing, meat quality

1. Introduction

Evolution of food processes is driven by changes in consumer preferences and the need to produce safe and high-quality foods. Nonthermal or intermediate technologies have great potential to achieve the characteristics desired by both the industry and consumers, especially regarding the desire to avoid altering the flavor or nutritional content during production. These technologies, which include the use of high pressure, electrical pulses, microfiltration,

and ultrasonication, are especially designed for economy, simplicity, and energy efficiency. Ultrasound is an acoustic energy [1], and therefore, it is a nonionizing, noninvasive, and non-polluting form of mechanical energy [2]. These properties lead to a wide range of applications in the food industry. It is considered an emerging method with a great potential to control, improve, and accelerate processes without damaging the quality of food and other products [3, 4]. A low-power and high-frequency method is used to monitor the composition and physicochemical properties of food components and products during processing. Therefore, it contributes to control the properties that improve food quality. In recent years, research studies have been focused on assessing the effects of ultrasound on processes including mass transfer or marinating, meat tenderizing, crystallization, freezing, drying, degassing, filtration, foam production and reduction, emulsification, homogenization, and inactivation of microorganisms and enzymes [2]. Ultrasound has also been employed to optimize physicochemical characteristics, preparation processes for meat products, microbiological content, and sensory characteristics in fresh and processed meat [5]. Although ultrasound waves have been used to improve a wide variety of characteristics for a variety of matrices and processes, the appropriate conditions for scaling ultrasonic methods up to industrial levels have been established for a relatively small number of processes [4].

As emphasized by Chemat et al.[6], a key goal of ultrasound research is to study and analyze both desirable and undesirable degradation phenomena in foods resulting from ultrasonic treatment (e.g., ultrasonic processing may affect the texture and chemical composition of foods). For this reason, many research questions in the meat sector are yet to be elucidated. Although multiple reports have been published, still inconsistent results have been reported, maybe because of the specific nature of meat tissues and various factors of ultrasound application possibly involved in, affecting food properties. This review aims to identify the effect of ultrasound on the major quality characteristics of fresh meat. We believe the results will help establish a methodology to enable the scaling-up of ultrasonic technology to the industrial level.

2. Ultrasound overview

Ultrasound is a form of energy generated by a longitudinal mechanical wave whose vibration frequency is greater than 20,000 cycles per second (20 kHz), which is above the audible limit for humans. Sound is considered a pressure wave with one-dimensional propagation. The speed of an ultrasonic pulse depends on the acoustic properties of the medium, and the speed of sound propagation is greater in solids than in liquids and higher in liquids than in gases [7]. In an ultrasound system, the electrical energy is transformed into vibrational energy, which is mechanical energy [8] that has been transmitted through a sonicated medium. Part of the input energy is lost through conversion to heat, and the rest can produce cavitation. A fraction of the cavitation energy produces chemical, physical, or biological effects, while other fractions are reflected and consumed in the reemission of sound. Ultrasound ranges from 20 kHz to 10 MHz and is divided into three categories: (1) high-power (>5 Wcm^{-2}or 10–1000 Wcm^{-2}) and low-frequency (20–100 kHz); (2) medium-power and intermediate-frequency (100 kHz–1 MHz); and (3) low-power (<1 Wcm^{-2}) and high-frequency (1–10 MHz) [9]. Three different methods

are used to apply ultrasound to products: (a) direct application; (b) coupling to the device, and (c) immersion in an ultrasound bath [2].

Low-intensity, high-frequency ultrasound has analytical applications that provide information about the physicochemical properties of foods such as composition, structure, and condition [10]. Furthermore, unlike conventional analytical techniques, it is noninvasive and nondestructive [11] and the measurements are fast, automated, and easy to use in both laboratories and production lines. High-power ultrasound, also known as high-intensity ultrasound, may cause changes in the physical, chemical, or mechanical properties of foods. In the field of biochemistry, ultrasound was initially used to rupture cell walls, releasing their contents. Subsequent studies showed that high-power ultrasound can be used to activate the immobilized enzymes by increasing the rate of transport of substrates to enzymes [12].

3. Power ultrasound

Power or high-intensity ultrasound has emerged as a new and complementary technology with a high number of potential applications. Its effects are primarily mechanical: alternate cycles of expansion and compression are produced, causing the growth or formation of new bubbles in the medium [13]. When they reach a volume in which they can no longer absorb more energy, the bubbles implode violently, causing microcurrents and the collapse of liquid molecules, a phenomenon known as cavitation. The quantity of energy released by the cavitation depends on the kinetics of the bubble's growth and collapse. This energy increases with increasing surface tension at the bubble interface and decreases with increasing vapor pressure of the liquid [14]. The results of ultrasound in liquid media depends on variables such as the characteristics of the treatment medium (viscosity, surface tension, vapor pressure, nature and concentration of dissolved gas, presence of solid particles, and temperature), efficiency of the ultrasound generator (frequency and input power), and the size and geometry of the treatment container [15].

4. Applications in food

Ultrasound has potentially a wide range of applications in the food industry. Researchers have identified various areas in which ultrasound can be used effectively, such as in the modification and control of crystallization processes, liquid food degassing, enzyme inactivation, drying, filtration, and oxidation induction. [12].Ultrasound methods have also been used in emulsion preparation in fruit and vegetable dehydration, enzyme inhibition, microbial inactivation, and crystallization of fats and sugars. Another example of a successful application of ultrasound technology is acoustic drying. It can be performed at lower temperatures than those used in conventional methods because the heat transfer between a solid surface and a liquid surface increases by approximately 30–60%, reducing the probability of oxidation or degradation of the material [15]. In addition, studies have shown that ultrasound is an effective method for food freezing, and the acceleration of ice nucleation and freezing leads to better control of the crystal size distribution in frozen products [16]. Ultrasound can not only

increase the speed of freezing fresh foods, but also improve the quality of frozen products. Currently, the pasteurization and conventional thermal sterilization are the most commonly used techniques for removing the threat posed by microorganisms in food products. Heat treatment destroys the vegetative microorganisms and some spores; however, its effectiveness depends on the treatment temperature and time. The magnitude, temperature, and time of treatment are also proportional to nutrient loss, development of undesirable flavors, and deterioration of functional properties of food products [17, 18]. Studies have shown that high-power ultrasound substantially reduces microbial loads because cavitation disrupts cell walls, resulting in the destruction of living cells and thereby contributing to food preservation. Unfortunately, scarce is still known about the mechanism of inactivation.

5. Applications in meat

The use of ultrasound in the meat industry, which began with the evaluation of live cattle fat and muscle, has been conducted since the 1950s. Nowadays, low-intensity ultrasound is routinely used to improve quality, taste, and tenderness, which represent the most important quality attributes in consumer satisfaction.

Many recent studies have reported potential uses of high-intensity ultrasound on fresh meat. Applications have been published with interesting advantages in freezing [19], thawing [20], meat brining [14], cooking [2], bacterial inhibition [21], and tenderizing [22]. The resulting changes of the application of ultrasound to fresh meat are summarized in **Table 1**.

Sample	Application (intensity/ freq/time)	Effect of ultrasound	Authors
Beef (*longissimus thoracis* and *lumborum,* and *semimembranosus*)	62 W cm^{-2}, 20 kHz, 15 s	No effect on mastication force, sensory traits, solubility of collagen or myofibrillar proteolysis.	Lyng et al. [58]
Semimembranosus pre- and post-rigor	10 W cm^{-2}, 2.6 MHz, 2 x15 s	Larger sarcomeres, Z-line disruption, increased calcium. No effect on collagen.	Got et al. [30]
Beef (*semimembranosus*)	2 W cm^{-2}, 25 kHz, 1 or 2 min	Lower loss of water after cooling, thawing, and heating. No effect on pH. Higher water holding capacity.	Dolatowski et al. [32]
Beef (*semimembranosus*) matured for 24, 48, 72 or 96 h at 2°C	2 W cm^{-2}, 45kHz, 2 min	No effect on meat color. Increased free calcium. Changes in protein structure. Improved WHC at 4 d *postmortem*.	Dolatowski and Stadnik [16]
Beef (*semimembranosus*) 24 h postmortem and matured for 24, 48, 72 or 96 h at 2°C	2 W cm^{-2}, 45kHz, 2 min	No effect on pH or color. Reduced hardness.	Stadnik and Dolatowski [35]

Sample	Application (intensity/ freq/time)	Effect of ultrasound	Authors
Beef (*semimembranosus*) 24 h postmortem and matured for 24, 48, 72 or 96 h at 2°C	2 W cm^{-2}, 45 kHz, 2 min	Acceleration of aging process. Fragmentation of protein structures. Increase WHC.	Stadnik et al. [31]
Beef longissimus lumborum et thoracis and semitendinosus aged up to 8.5 days	12 W cm^{-2} , 24 kHz, for up to 240 s	Reduced WBS force and hardness. Increased pH. No interaction between ultrasound and aging. No changes in meat color and drip loss. Ultrasound reduced cook and total loss.	Jayasooriya et al. [34]
Hen breast meat stored for 0, 1, 3, or 7 d at 4°C	12 W cm^{-2}, 24 kHz, 15 s period	Reduced shear force. No change in cooking loss.	Xiong et al. [53]
Beef (*semitendinosus*)	1500 W, 40 kHz, 10, 20, 30, 40, 50, or 60 min	No effect on brightness and red color. Decreased the tendency to yellow. Decreased the muscle fiber diameter. No effect on heat-insoluble collagen. Weaken collagen stability.	Chang et al. [61]
Pork *biceps femoris* *24 h post mortem*	150 W, 1 MHz and 500 W, 25 kHz, 40 min	Ultrasound did not change in shear force. Ultrasound combined with actinidin decreased shear force more than actinidin alone.	Jørgensen et al. [60]
Beef (*semimembranosus*) 24 h postmortem and matured for 24, 48, 72 or 96 h at 2 °C	2 W cm^{-2}, 45kHz, 2 min	Slightly less stable color. No change in oxidative stability at 4 d storage.	Stadnik et al. [31]
Beef *semitendinosus*	40 kHz, 11 w cm-2 for 0, 60 and 90 min.	Increases luminosity and reduces redness up to 8 d of storage. No effect on water holding capacity of meat. Decreased coliforms and psychrophilic bacterial load.	Caraveo et al. [21]
Beef *longissimus thoracic* and deep *pectoralis* Matured 14 d at 2°C Cooked at 62°C or 70°C	1000 W, 20 kHz, 0, 5 or 10 min	Faster cooking, higher water retention, decreased cooking loss, shear force and soluble collagen. Higher sensory tenderness.	Pohlman et al. [44]
Holstein bulls (*longissimus lumborum)*	20 kHz, 100 and 300 W for 10, 20 or 30 min	Improved meat tenderness, decreased shear force, filtering residue and textural parameters.	Barekat and Soltanizadeh [55]

Sample	Application (intensity/ freq/time)	Effect of ultrasound	Authors
Beef *longissimus dorsi*	40 kHz, 11 W cm-2 for 60 min.	Reduces shear force. Produces more tender and juice meat. No effect on meat color.	Peña-González et al. [22]
Chicken breast and soybean gels, 4°C to 8°C	450 W, 20 kHz, 0, 3, 6, 9 or 12 min (4 or 2 s pulses)	More viscoelastic gel Improved WFB and textural properties Homogeneous fine network microstructures	Zhao et al. [54]
Chicken breast	22 W cm^{-2} 40 kHz, 15 or 30 min	Increased mass transfer and higher meat weight	Leal-Ramos et al. [62]
Pork *longissimus dorsi*	100 W and 20 kHz, 45 min	Increased salt gain and water loss. Higher mass transfer at higher ultrasound intensity.	Cárcel et al. [14]
Pork *longissimus dorsi*	2-4 W cm^{-2}, 20 kHz, 30, 90 or 180 min	Higher salt diffusion. Diffusion coefficient increases with ultrasound intensity.	Siró et al. [15]
Pork *longissimus dorsi*	40 kHz; 37.5 W dm^{-3}, 15, 30, 45, 60, 90 or 120 min	Higher salt and water diffusion.	Ozuna et al. [63]
Pork *longissimus thoracis and lumborum*	4.2, 11 or 19 W cm^{-2}, 20 kHz, 10, 25 or 40 min	No effect on water holding capacity and structure of meat. Higher mass transfer and protein extraction. Myosin denaturation at higher intensities.	McDonnell et al. [59]
Pork meat and skin surface	High-intensity ultrasound, 0.5 to 2 s	Less skin and surface bacteria	Morild et al. [80]
Chicken breast	Ultrasonic bath, 20 min	No effect on water retention capacity, shear force and cooking loss. No changes in *Salmonella* and *E. coli*.	Smith et al. [43]
Chicken wing surface	2.5 W cm^{-2}, 40 kHz, 3 or 6 min	Microorganism reduction. Higher reduction with higher time. *E. coli* more sensible to ultrasound.	Kordowska-Wiater and Stasiak [82]
Pure culture suspensions	20 kHz, 3, 6 or 9 min, 20, 40 and 60 °C	Bacteria inactivation is higher at higher time and temperature.	Herceg et al. [84]
Chicken carcasses	SonoSteam®	*Campylobacter* and viable total count reduction.	Musavian et al. [83]

Table 1. Effects of ultrasound on meat.

6. Effects on meat quality properties

The majority of quality parameters assessed 24 h *postmortem* in meat is physicochemical in nature [23]. The potential of hydrogen (pH), water holding capacity (WHC), drip loss (DL), color (L*, a*, b*, C*, and h*), and shear force are quality indicators for the raw meat. Physicochemical characteristics are evaluated to control the quality, assess the efficiency of production and treatment processing, compare results between laboratories, and aid research [24, 25]. The effect of a number of nutritional, breed, and production factors such as genetics, management, and slaughter on specific quality attributes should be considered when meat quality is analyzed [26]. Key markers of meat quality, including raw materials, which have a strong impact on the industry, are pH, water holding capacity, texture, and oxidative stability. In addition, aroma, color, flavor, and tenderness are the most important sensory components to the meat-consuming population [27]. When considering the use of emerging technologies in meat, both the mechanisms of action and the effects on food transformation, preservation, and integrity must be known. Ultrasound application may be an efficient alternative technology to increase meat tenderness. It is used in meat processing and preservation as a complementary or assistive technology [28].

6.1. Potential of hydrogen (pH)

pH is one of the most important indicators of raw meat quality, because it directly affects protein stability and properties. As reported in the literature, all product quality attributes depend on the ultimate pH. Water holding capacity, drip loss, and color are among the most important product quality attributes [29]. pH greatly affects the quality and functionality of muscle proteins, and decrease in pH reduces their water holding capacity, leading to economic losses. On the other hand, increases in pH increase the water holding capacity of the meat because of changes in the electrical charges within muscle proteins that occur when the pH is above the isoelectric point. Ultrasound effect on pH has been analyzed in various studies and conditions. Some authors have reported that initial pH of meat can be increased as a result of ultrasound treatment (2.6 MHz, 10 W/cm^2) [30] prior to *rigor mortis*, with no differences in final pH, while other reported small differences [31] or no differences in pH because of ultrasound [32–35].In a study, bovine muscles (*longissimus lumborum et thoracis* and *semitendinosus*) sonicated (24 kHz, 12 Wcm^{-2}) for a maximum of 4 min and subsequently stored them for 8 days led to increase tenderness and pH without a significant correlation between ultrasound and aging time [34]. The increase of pH was attributed to the release of ions from the cellular structure into the cytoplasm or to changes in protein structure, which could lead to changes in the position of ionic functionalities that could lower the muscle pH.

6.2. Water holding capacity (WHC)

Water holding capacity (WHC) may be defined as the ability of meat or muscle proteins to immobilize their own or added water during an applied force [25]. A decrease in pH causes a shrink of the network of polypeptide chains, which decreases the water holding capacity.

Therefore, WHC is directly related to pH. The speed to reach the final pH also affects the WHC. When the drop in pH is relatively rapid, the changes in myofibrillar and sarcoplasmic proteins result in a decreased water holding capacity [29]. Some meat characteristics linked to WHC include color, texture, firmness, juiciness, and tenderness. Meat WHC is affected by factors including *rigor mortis*, ATP loss, and changes in the myofibrillar structure partly associated with proteolytic activity. Many other characteristics, including drip loss, are closely related to or depend on WHC.

Approximately, three-quarter of meat is water, and about 10% of the water in live animal muscle is bound to muscle proteins compared to the 5–10% of water located in small channels between adjacent cells, or extracellular space. However, most of the water content is located in spaces between thin and thick filaments of myofibrils. In any muscle, WHC is minimal at low pH. Because of aging, it tends to increase owing to protein degradation and changes in electric charges induced by intramolecular reorganization [23]. When the WHC is low, moisture or weight loss during storage is greater owing to surface evaporation and exudation of cuts because the WHC is related to several physicochemical characteristics of the protein and myofibrillar components. WHC is the main indicator of the suitability of a given meat for preparing a product. The effects of ultrasonic treatment on WHC of meat are variable. Variation of effects on WHC is described on the next section, due to the relation between WHC and drip loss.

6.3. Drip loss (DL)

The release of water droplets from the muscle originated from the extracellular water is known as DL. It is the easiest water content to extract. DL depends on the state of contraction after *rigor mortis* because of reduction in the filamentary space and changes in the cellular membrane, which causes the release of water to the extracellular space in the form of drops through the cutting surfaces [35]. These drops consist of an aqueous red solution that largely contains proteins and water-soluble minerals, some of which are highly nutritious. Drip loss is strictly related to pH and WHC. When WHC increases, DL decreases and vice versa [36]. Several factors increase the WHC during meat aging, including pH, Z-line disintegration by protease activity, and changes in membrane permeability with diffusion and ionic redistribution, which results in substitution of divalent ions and weakening of intermolecular forces between protein chains. DL is primarily an economic problem for retailers, because weight losses during cutting cause accumulation of liquid around the product, which leads to consumer rejection [37, 38]. WHC is a key indicator of meat quality that affects the economic sector. Therefore, analyzing the effect of ultrasound on WHC is important [39]. Assessment of DL is used to identify the best conditions for the refrigeration, freezing, packaging, and storage conditions of meat. Consequently, DL measurements also make possible to determine WHC. The effects of ultrasonic treatments on WHC and drip loss are highly variable. Some authors report that ultrasound increases the rates of meat exudation and water loss [40, 41]. Whereas, other authors found no effect on the water holding capacity [42] or drip loss [21, 34] in beef (24–40 kHz, 11–12 W cm^{-2}). In contrast, some reports indicate that ultrasonicated meat has a higher WHC [16, 40, 43], similar to that of meat at an advanced *postmortem* stage. This could be explained by structural changes in myofibrillar proteins caused by ultrasound; the above is confirmed by microstructure photographs [31]. Recently, when high-intensity ultrasound is applied during brining of beef, higher WHC was found, possibly by a higher diffusion of salt into the tissue, which can increase the capacity to hold the water before cooking

the meat [44, 45]. The variability seen in the literature could result from differences in the ultrasound methods; the authors employed various times and intensities, which hindered direct comparison.

6.4. Color

Color is a key factor in meat quality because it is the first sensory characteristic assessed by the consumer [46]. In red meat, a bright red color is related to freshness and therefore consumer rejection or acceptance [47]. Meat color results from the quantity and chemical state of myoglobin in the muscle. Deoxymyoglobin and myoglobin are responsible for the purple color of fresh meat. When meat is exposed to air for several minutes, deoxymyoglobin is oxygenated into oxymyoglobin, which is responsible for a cherry red color in meat. When meat is exposed to air for several hours or days, it turns brown due to the oxidation of oxymyoglobin into metmyoglobin. Meat contains other pigments, some derived from external sources, sometimes in insignificant amounts, which commonly indicate deterioration. Meat color and exterior appearance may be associated with aging time, shelf life, hardness, and juiciness. Some studies suggest that ultrasound has no effect on meat color because the heat generated is insufficient to denature proteins and pigments [48, 49]. Conversely, in an assessment of the effect of ultrasound (22 W/cm^2) on meat, it has been found that the color changed to a lighter, less red, and more yellow-orange color (greater hue angle), which was less bright than control meat [43]. Ultrasound accelerates total changes in color, limits the formation of oxymyoglobin, and slows down the formation of metmyoglobin [48]. Nevertheless, when meat is cooked, meat panelists do not detect differences between ultrasonicated and control meat [22].

6.5. Tenderness

Tenderness in meat is determined by its texture. Tenderness is one of the most important attributes of meat quality because it is perhaps the most appreciated feature by consumers. Inconsistencies in this characteristic have been considered one of the major problems that the meat packing industry faces [50]. Tenderness is affected by the composition, structural organization, and integrity of the skeletal muscle. The two structural components that determine the intrinsic muscle strength are myofibrillar proteins and connective tissues [51] and the nature of these two components makes difficult to achieve tenderness. Tenderness depends on the size of the longitudinally arranged fiber bundles in muscles, which are delimited by the connective tissue septa forming the perimysium [25]. Myofibrillar tenderness can be controlled by manipulating conditions *pre-* and *postmortem*. Some methods and procedures used to increase tenderness include electrical stimulation, pressurization, calcium infusion, enzymatic treatment, and marination. All these methods are invasive, cause deformation, and affect the appearance of meat. In addition, some methods may contaminate the meat (e.g., brine injection with unclean needles). Currently, aging is the foremost industrial process used to increase the tenderness of meat. Aging tenderization mechanism is well known nowadays, consisting of biochemical processes driven by endogenous proteases. Nevertheless, aging is a time-consuming process, and it can be variable among animals. Therefore, various physical methods, such as electrical stimulation and chemical methods, have been used trying to improve tenderness.

Numerous studies have been conducted to develop methods to improve tenderness. Among these, ultrasound application methods have been used at various sonication times, frequencies, and intensities. Most authors agree that ultrasound increases meat tenderness [22, 40, 48] and shortens the aging period without compromising other quality parameters [16, 33]. The potential of low-frequency, low-intensity ultrasound application to improve meat tenderization is remarkable. Several authors report an important reduction of shear force after treatment with ultrasound [52]. Benefits of ultrasound treatment on beef have been observed in *longissimus lumborum* and *semitendinosus* (24 kHz and 12 W/cm^2 for 240 s) [34], *M. Semimembranosus* (45 kHz and 2 W/cm^2 for 2 min) [48], and *semitendinosus* (40 kHz, 1500 W for 10, 20, 30, 40, 50 or 60 min) [40]. Benefits to the texture of poultry (24 kHz, 12 W/cm^2 for 4 min after 7 d of storage) [53, 54] and pork (2.5–3 W/cm^2 for 180 min) [15] are also reported. It has also been observed a significant decrease in the shear force of *Bovine L. dorsi* with the application of ultrasound, both, fresh and aged [22]. More recently, it was reported a reduction of shear force values in muscle semitendinosus when it was ultrasonicated and aged for 3–7 d [45]. The effect was mainly attributed to an increase of desmin and troponin-T degradation, and myofiber fracture along Z-lines and I-bands.

It is suggested that acoustic cavitation may induce mechanical rupture of myofibrillar protein structures [31], fragmentation of collagen macromolecules, migration of proteins, minerals and other compounds, thereby accelerating proteolysis or protein denaturation. High-intensity ultrasound can cause degradation of cells and some subcellular components, because periodic oscillation of acoustic pressure softens cell membranes. Research has also evinced tissue disruption in the migration of proteins, minerals, and other components; accelerating enzymatic activity and degradation of collagen macromolecules when meat is exposed to high-intensity ultrasound [11, 55]. In addition to tenderize, high-intensity ultrasound can also improve meat sensory properties [22]. After applying ultrasound, the quantity of ATP available in muscles at pre-*rigor* stage may change [49], accelerating the start of *rigor mortis* [56]. Indirectly, ultrasound may induce tenderization because of the activation of proteolysis by the release of lysosome cathepsins and/or intracellular calcium ions that activate calpains. This mechanism may lead to a weaker cellular structure [48] through protein denaturation, which in turn causes muscle tissue disruption that results in increased tenderness [40] and a shortened aging period [57]. It should be noted that some reports also indicate that ultrasound has no effect on shear force when it is applied at 62 W/cm^2 [58], 22 W/cm^2 [43], 4–19 W/cm^2 [59], or 150 and 500 W [60]. The data available thus far indicate that ultrasound does indeed exert a key effect on meat tenderization, although the application parameters must be established before the method can be scaled to industrial levels. The effect of ultrasound on the physicochemical characteristics and *semitendinosus* muscle collagen has been studied [61]. Their results suggest that ultrasound affects denaturation and aggregation of collagen fibers in the extracellular space. These changes contribute to benefit the quality and texture of the meat. Besides, meat luminosity and tendency to redden remained unaffected.

6.6. Marination to improve meat quality

High-intensity ultrasound application during meat marination has been frequently studied. Meat marinades may contain salt in two forms: dry or wet [14]. High-intensity ultrasound application resulted favorable for salt diffusion when used in wet marinades. The effect

of power ultrasound on pork during wet marination depends on the ultrasound intensity applied [62]. Ultrasound causes bubble formation that hits the tissue, which may lead to microinjection of brine into the sample. This effect may help to explain the observed increase of NaCl content in the ultrasonicated meat [14, 63].

Ultrasonic treatment (low-intensity and low-frequency) and the use of vacuum caused favorable microstructural changes in pork loins marinated in sodium chloride [15] and these effects are highly dependent on the intensity of ultrasound treatment. Some of the critical factors in food processing warrant consideration because ultrasound generates rapid changes in temperature and pressure (109°C/s) over short time periods. Furthermore, cavitation generates shock waves, which contribute to this effect. Factors that modulate the effects of ultrasound application include time of exposure, processing volume, and sample composition [12, 14].

6.7. Microbiological properties

Bacteria are the most important microorganisms in food processing. While most are harmless and many are beneficial, some indicate the likely presence of contamination and deterioration and may cause diseases. While thousands of bacterial species have been identified, all are unicellular and fall under three basic forms: spherical, rod-shaped, and spiral. Some rod-shaped bacteria can take two forms: latent spores and active vegetative cells. The vegetative cells form spores under adverse conditions to survive. Most sporulating bacteria that grow in the presence of air belong to the *Bacillus* genus, and most of those that grow only in the absence of air belong to the genus *Clostridium*.

Meat is susceptible to the growth of some pathogenic microorganisms such as *E. coli, Campylobacter jejuni, Salmonella spp., Staphylococcus aureus,* and *Listeria monocytogenes*, which recurrently affect the properties of the meat and present serious problems during packing, processing, and storage. Several methods are used to avoid microbial growth in meat. The most commonly used methods involve heating, dehydration, and addition of preservatives [64]. The most common types of mesophilic bacteria that are pathogenic to humans include *Staphylococcus aureus*, *Salmonella*, and *Listeria*. Although it may survive without damage in the intestinal tract of humans, salmonella is a common cause of food poisoning. Another common mesophilic bacterium, *Listeria monocytogenes*, is more often distributed through contaminated foods such as raw meats or unpasteurized cheeses [64]. Animals, including humans, may transport *Listeria*, but it primarily threatens those with weakened immune systems. Some *E. coli* strains found in human feces are pathogenic, causing infection and disease. These are called enteropathogenic bacteria.

Staphylococcus is nonsporulated bacteria without mobility, but because they are resistant to drying, they are easily dispersible by dust particles through air and surfaces [65]. *S. aureus* is usually found in the skin and in mucous membranes of humans and other animals. It is almost always present in small quantities in raw meats and foods extensively handled by humans. Maintaining food that is completely free of contamination with *Staphylococcus* is often difficult or impossible. Pasteurizing or cooking destroys the organism but not its toxin [66].Meat is one of the most perishable foods consumed by humans—it is easily damaged by bacteria. One of the most commonly used preservation methods is refrigeration, including freezing. However, certain bacteria are able to grow at 4°C; these are collectively known as psychrophiles. This

group includes some pathogens such as *Yersinia enterocolitica*, *Listeria monocytogenes*, non-proteolytic strains of *C. botulinum*, and some strains of enterotoxigenic *E. coli* and *Aeromonas hydrophila*. Several other organisms that can cause foodborne diseases and grow at refrigeration temperatures include: *Vibrio parahaemolyticus*, *Bacillus cereus*, *Staphylococcus aureus,* and some *Salmonella* strains [64].

When refrigeration is extended, *Pseudomonas*, *Acinetobacter*, and *Moraxella* species may grow and damage fresh meat [67]. Gram-negative organisms are known to survive less frequently compared to their Gram-positive counterparts [68–70]. However, recent studies have shown higher survival rates among Gram-negative bacteria, especially *Pseudomonas* species, which account for the majority of bacteria responsible for refrigerated meat deterioration [67]. Refrigerated foods, such as processed meat, should be stored as close as possible to 0°C. However, in most cases, they remain close to 4–8°C. This fluctuation in temperature reduces the useful life of the products and can lead to major public health problems. The fresh meat industry must incorporate as many treatments as possible that reduce the microbial population and minimize reproduction. Some of these treatments include heat, acidification, preservatives, reduced water activity, and packaging under modified atmospheres. Although modified atmospheres are included as a potential barrier, it should be noted that reduced oxygen atmospheres can actually favor anaerobic pathogens. For many products, the modified atmosphere actually helps improve product quality rather than safety.

Yeast and molds grow on most foods, equipment, and building surfaces with small amounts of nutrients and moisture [71]. Because bacteria grow faster, they greatly outgrow yeasts and molds in most foods. Fungi and yeasts grow well in low-pH, humid, and temperature environments with high concentrations of salt and sugar. Therefore, they can pose a problem in dry foods, such as dried meat and salted fish [72].

Effective microbial destruction is of paramount importance for food processing; a single report of microbial contamination could question the reputation of a manufacturer and jeopardize their future success. To minimize the bacterial load of a product, the manufacturer must reduce the initial contamination, inactivate microorganisms present in the food, and implement procedures to prevent or slow the growth of microbial populations that have not been inactivated. Conventional methods of bacterial inactivation involve thermal treatments, such as pasteurization. These treatments generally result in undesirable flavors and the loss of nutrients. Ultrasonic treatment has been used to inactivate bacterial populations [73]. This is due to cavitation effects: pressure changes produced by the ultrasonic waves cause microbiological inactivation [3, 73]. The microbiological damage resulting from the application of various ultrasound wave amplitudes depends on factors such as contact time with the microorganism, microorganism type, food quantity, composition, and treatment temperature [74]. Microbial resistance varies among microorganisms, i.e., some are more susceptible than others to the ultrasound process. Studies have shown that larger or longer cells are more susceptible to ultrasound because they have more a larger contact surface and are therefore more exposed to the pressure produced by cavitation [75]. Gram-positive bacteria are less susceptible to ultrasound compared to Gram-negative bacteria, although results have shown that rod-shaped (bacillus) microorganisms tend to be more susceptible than cocci [76].

Gram-positive bacteria are likely less susceptible to ultrasound because of their thicker cell walls, which contain an adhesive peptidoglycan layer [77, 78]. In general, microorganisms that produce spores exhibit a greater resistance to heat and ultrasound [74, 75].

A considerable amount of data on the impact of ultrasound on microbial inactivation is available. One study demonstrated the effects on the microbiological environment of bacterial suspensions by inoculating the skin of broilers with *Salmonella*; the Salmonella population decreased with ultrasound treatment in peptone at 20 kHz for 30 min [16]. Studies have shown that the intensity of traditional heat treatments can be reduced by 50% when they are combined with power ultrasound. For this reason, a new method for antimicrobial treatment could feature the combined effects of pressure and ultrasound (manosonication), ultrasound and heat (thermosonication), or ultrasound, heat, and pressure (manothermosonication) [79]. These are likely the best microbial inactivation methods because they are more energy-efficient and effective in inhibiting microorganisms than conventional methods. The effectiveness of ultrasonic treatments requires prolonged exposure to high temperatures, which may deteriorate functional properties, sensory characteristics, and the nutritional content of foods [73]. In combination with heat, ultrasound can accelerate the rate of food sterilization, thereby decreasing the necessary duration and intensity of heat treatment and the resulting damage.

The inactivation of *Salmonella typhimurium, Salmonella derby, Salmonella infantis, Yersinia enterocolitica,* and a pathogenic strain of *Escherichia coli* was studied in inoculated samples treated for 0.5–2.0 s. The total viable bacterial counts decreased by 1.1 log CFU cm^{-2} after a 1-second treatment and by 3.3 log CFU cm^{-2} after a 4-second treatment [80]. The reduction of the population in the skin was significantly greater than that in the meat, although no significant differences were observed between the types of bacteria. However, the study by Smith *et al.* [81] stands out. They reported no effect after ultrasound on *Salmonella* or on *E. coli* in marinated chicken, likely because ultrasound alone is not fully effective in bacterial inhibition.

Some authors [82] have studied the elimination of Gram-negative bacteria (*Salmonella anatum, Escherichia coli, Proteus* sp., and *Pseudomonas fluorescens*) on the surface of chicken skin after ultrasonic treatment (40 kHz and 2.5 Wcm^{-2} for 3 or 6 min) in water and in 1% aqueous lactic acid. Sonication in water alone or lactic acid solutions for 3 min resulted in a decrease in the number of microorganisms on the surface of the skin of 1.0 CFU cm^{-2}. Other reports show that treating chicken carcasses in the process line with steam and ultrasonic treatments significantly reduces the population of *Campylobacter* in contaminated poultry. The total viable content decreased by approximately three logarithmic units when steam and ultrasound were applied immediately after slaughter [83]. Ultrasound treatments combined with lactic acid may be a suitable method for decontaminating poultry carcass skins.

Ultrasound effects depend on frequency, amplitude, time, and temperature [84] as it was demonstrated on the inactivation of suspensions containing *Escherichia coli, Staphylococcus aureus, Salmonella* sp., *Listeria monocytogenes*, and *Bacillus cereus* treated with a 12.7-mm ultrasound probe at 20 kHz and 60, 90, and 120 mm amplitudes for 3, 6, and 9 min at 20, 40, and 60°C. These three parameters affected the inactivation of bacteria in pure cultures. The results showed increased microbial inactivation for longer treatment periods, particularly when they were combined with high temperature and amplitude.

It has been observed that treating fresh beef with a power ultrasound method decreased its bacterial load, particularly of coliforms and psychrophilic bacteria, when a frequency of 40 kHz and intensity of 60 W/cm^2 were applied for 60 and 90 min. Meat treated for the longer period showed the largest reduction of microorganisms during storage [21].

7. Conclusions

Selected and potential applications of ultrasound mainly in the field of food preservation and product modification were discussed. High-intensity ultrasound generates acoustic cavitation in a liquid medium, developing physical forces that are considered the main mechanism responsible for the observed changes in exposed materials. These forces include acoustic streaming, cavitation, shear, micro-jet, and shockwaves. The quantity of energy released by the cavitation depends on many factors such as treatment medium and ultrasound frequency. Ultrasound has a wide range of applications in the food industry. It can be used as a processing aid in extraction, crystallization, freezing, emulsification, filtration, and drying. Applications of ultrasound in meat have been reported with interesting advantages in freezing, thawing, meat brining, and tenderizing. Ultrasound has also been shown to improve physicochemical characteristics, preparation processes for meat products, microbiological content, and sensory characteristics in fresh and processed meat. Acoustic cavitation may induce the mechanical rupture of the myofibrillar protein structure with significant effect on collagen characteristics and meat textural properties. High-intensity ultrasound reduces microbial loads in meat, resulting in the destruction of living cells and this effect remains during cold storage. Like most innovative food processing technologies, high-power ultrasonics needs to be developed and scaled up for each application.

8. Conflict of interest

The authors declare that they have no interest or benefit arising from the direct applications of this chapter.

Author details

Alma D. Alarcon-Rojo[1]*, Esmeralda Peña-González[1], Iván García-Galicia[1], Luis Carrillo-López[2], Mariana Huerta-Jiménez[2], Raúl Reyes-Villagrana[2] and Hector Janacua-Vidales[3]

*Address all correspondence to: aalarcon@uach.mx

1 Autonomous University of Chihuahua, Chihuahua, Mexico

2 CONACYT - Autonomous University of Chihuahua, Chihuahua, Mexico

3 Autonomous University of Ciudad Juarez, Chihuahua, Mexico

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